Translation

PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference 432725GA	FOR FURTHER ACTION	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
International application No. PCT/EP2003/006566	International filing date (*day/month/year*) 23 June 2003 (23.06.2003)	Priority date (*day/month/year*) 28 June 2002 (28.06.2002)
International Patent Classification (IPC) or national classification and IPC G01N 33/543		
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1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 7 sheets, including this cover sheet.

☒ This report is also accompanied by ANNEXES, i.e., sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

These annexes consist of a total of 4 sheets.

3. This report contains indications relating to the following items:

- I ☒ Basis of the report
- II ☐ Priority
- III ☐ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
- IV ☐ Lack of unity of invention
- V ☒ Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
- VI ☐ Certain documents cited
- VII ☐ Certain defects in the international application
- VIII ☐ Certain observations on the international application

Date of submission of the demand 28 November 2003 (28.11.2003)	Date of completion of this report 30 September 2004 (30.09.2004)
Name and mailing address of the IPEA/EP Facsimile No.	Authorized officer Telephone No.

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/EP2003/006566

I. Basis of the report

1. This report has been drawn on the basis of (*Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to the report since they do not contain amendments.*):

☐ the international application as originally filed.

☒ the description, pages 1-14, as originally filed,
pages _____, filed with the demand,
pages _____, filed with the letter of _____,
pages _____, filed with the letter of _____.

☒ the claims, Nos. _____, as originally filed,
Nos. _____, as amended under Article 19,
Nos. _____, filed with the demand,
Nos. 2, 4, filed with the letter of 12 August 2004 (12.08.2004),
Nos. 1,3, filed ~~with the letter of~~ Personally.

☒ the drawings, sheets/fig 1,2, as originally filed,
sheets/fig _____, filed with the demand,
sheets/fig _____, filed with the letter of _____,
sheets/fig _____, filed with the letter of _____.

2. The amendments have resulted in the cancellation of:

☐ the description, pages _____
☐ the claims, Nos. _____
☐ the drawings, sheets/fig _____

3. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).

4. Additional observations, if necessary:

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No.
PCT/EP 03/06566

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Claims	1-14	YES
	Claims		NO
Inventive step (IS)	Claims	1-14	YES
	Claims		NO
Industrial applicability (IA)	Claims	1-14	YES
	Claims		NO

2. Citations and explanations

1.1 This report makes reference to the following documents:

D1: US 4 655 880 (cited in the application)

D2: US2001/0029048 A1

D3: US 5 149 629 (cited in the application)

D4: US 4 315 753 (cited in the application)

D5: US 5 217 112

1.2 **Novelty - independent claims 1 and 12**:

D2, which is regarded as the closest prior art, discloses a device (see figures 1 and 2) for the electrochemical detection of at least one type of biochemical molecule contained in a liquid from a group of predetermined biochemical molecules of different types (paragraph 1, lines 1 and 2), said device having at least one reference electrode 26 (paragraph 9, line 9) and at least one counter electrode 28 (paragraph 9, line 9) and more than two working electrodes 22, 24 (paragraph 9, lines 7 and 8, paragraph 4, paragraph 38, lines 8 to 11) having means 10, 12 for receiving the liquid (paragraph 9, lines 1 and 2).

The following features of claim 1 (and the corresponding features of claim 12) are not disclosed

in D2:

- "for detecting each type of biochemical molecule, at least one working electrode is provided which is coated with a molecule that is complementary to the biochemical molecule to be detected" (in D2, said molecules are not arranged on the corresponding working electrode, but on a substrate next to the working electrode at a certain distance, see paragraph 38);
- "a potentiostat for generating a predetermined voltage curve between the working electrodes and the reference electrode, this voltage curve being alterable during the measurement," (in D2, a separate potentiostat is used for each working electrode, see paragraph 24; the applied voltage is kept constant, see paragraph 26);
- "a current-voltage converter is connected downstream of each of the working electrodes, the current-voltage converters keep all of the working electrodes at the same potential" (current-voltage converters are not mentioned in D2).

D1 discloses (see, in particular, figure 6 and the corresponding description) a device of the aforementioned type for the electrochemical detection of a biochemical molecule contained in a liquid, only two working electrodes being provided, of which only one is correspondingly coated; the other one is used for background compensation. Although the voltage between the working electrodes and the reference electrode is generated by only one potentiostat, this potentiostat keeps the applied voltage constant. A current-voltage converter is connected downstream of each of the two working electrodes.

D3 also discloses (see figure 1 and the corresponding description) a device of the aforementioned type for the electrochemical detection of a biochemical molecule contained in a liquid, said device having a plurality (example 8) of correspondingly coated working electrodes and only one potentiostat. The potentiostat keeps the working electrodes at a constant potential. Current-voltage converters connected downstream of the working electrodes are not disclosed (in order to be able to read out the working electrodes with only one potentiostat, these working electrodes are connected to a multiplexer and read out serially).

D4 discloses a device for simultaneously measuring NO_2 and NO, this device not being suitable for detecting a biochemical molecule in a liquid. The device has only two working electrodes that are not coated. The potentiostat used there keeps the working electrodes at a constant potential.

D5 discloses a device for detecting ions and gases in a liquid, this device not being suitable for detecting a biochemical molecule in a liquid. Although the device has a plurality (example 5) of working electrodes, these electrodes are not coated, but consist of different materials (for example, C, Au, Ag, Ni, Pt). A voltage curve that can be altered during the measurement is generated at the working electrodes with only one potentiostat, but, in contrast to the present application, the voltage curve is generated between the working electrodes and the counter electrode, not between the working electrodes and the reference electrode (see D5,

column 3, lines 11-17). Current-voltage converters connected downstream of the working electrodes are not mentioned.

Claims 1 and 12 therefore meet the requirement of novelty under PCT Article 33(2).

1.3 **Inventive step - independent claims 1 and 12**:

Proceeding from D2, the **problem addressed by the invention** is that of providing a device and a method with which simultaneous electrochemical detection of different biochemical molecules contained a liquid can be carried out with the lowest expenditure on apparatus possible and with the greatest accuracy possible.

The invention solves the problem by the combination of features in claim 1 and claim 12.

A person skilled in the art knows to coat electrodes correspondingly (D1, D3). A person skilled in the art also knows to use only one potentiostat for a plurality of working electrodes and to read out the working electrodes by means of current-voltage converters (D1). However, a person skilled in the art does *not* know, in connection with the detection of biochemical molecules, to generate a voltage curve that can be altered during the measurement at the working electrodes. In order to arrive at the subject matter of the application, a person skilled in the art would have to combine (at least) three documents (D2 + D1 + D5). This, however, would amount to an *ex-post-facto* analysis, since there is nothing in these documents that would suggest combining them. It is therefore uncertain whether a person skilled in the art of biochemical analytics would even consult D5, which relates to the detection of (inorganic) ions and gases. Even if a person skilled in the art were

to combine these three documents, there would still be an additional difference with respect to the claimed subject matter of the present application, namely that said voltage curve would have to be generated between the working electrodes and the reference electrode to be the same as the application.

Claims 1 and 12 therefore meet the requirement of inventive step under PCT Article 33(3).

1.4 **Claims 2 to 11 and 13 and 14** are dependent on claims 1 and 12, respectively, and therefore also meet the requirements of PCT Article 33(1).

2. For the sake of completeness, the following formal defects should be noted:

(i) the features "a second operational amplifier" (**claim 9**, claim 1 does not define a "first operational amplifier"), "a third operational amplifier", "a second resistor", "of the second operational amplifier" and "a third resistor" (**claim 10**) are not previously defined. These claims therefore appear to be unclear (PCT Article 6).

(ii) Moreover, the claims also appear to be unclear owing to contradictions in the description (see PCT International Preliminary Examination Guidelines, Chapter III, paragraph 4.3): page 12, lines 22-23, contradicts the present claims, since neither a circuit *per se* nor a device with only uncoated working electrodes is claimed.

(iii) Independent device claim 1 has not been drafted in the two-part form under PCT Rule 6.3(b).

(iv) Claim 13 should refer back to claim 12, not to itself.

(v) Contrary to PCT Rule 5.1(a)(ii), the description does not cite D2 or indicate the relevant prior art disclosed therein.

(vi) The description has not been brought into line with the amended claims (PCT Rule 5.1(a)(iii)).

(vii) Reference sign "1" for the "container" (see, for example, page 9, line 26) has not been included in the figures. This reference sign should therefore be deleted from the description.

New Patent Claims

1. A device for the electrochemical detection of at least one type of a biochemical molecule - contained in a liquid - from a group of predetermined biochemical molecules of different types, having
a means (1) for taking up the liquid, said means having at least one reference electrode (RE) and at least one counterelectrode (GE) and also more than two working electrodes (AE1, AE2, AE3), at least in each case one working electrode (AE1, AE2, AE3) being provided for the detection of each type of a biochemical molecule, said working electrode being coated with a molecule that is complementary to the biochemical molecule to be detected, so that biochemical molecules of different types can be detected simultaneously,
a potentiostat (P) for generating a predetermined voltage profile - which is variable during the measurement - between the working electrodes (AE1, AE2, AE3) and the reference electrode (RE),
a current/voltage converter (S1, S2, S3) being connected downstream of each of the working electrodes (AE1, AE2, AE3), the current/voltage converters (S1, S2, S3) holding all of the working electrodes (AE1, AE2, AE3) at the same potential and
a means (S1, S2, S3, AD) for measuring the currents flowing through the working electrodes (AE1, AE2, AE3).

2. The device as claimed in claim 1, a plurality of interconnected or capacitively coupled reference electrodes (RE) being provided.

3. The device as claimed in claim 1 or 2, a plurality of interconnected counterelectrodes (GE) being provided.

4. The device as claimed in one of the preceding claims, the measuring means (AD) having an analog-to-digital converter.

5. The device as claimed in one of the preceding claims, the current/voltage converter (S1, S2, S3) being a current follower having a first operational amplifier (OP1), a noninverting input (OP1+) of the first operational amplifier (OP1) being grounded and the inverting input (OP1-) thereof being connected via a first resistor (R1) to the output of the first operational amplifier (OP1) and to the working electrode (AE1).

6. The device as claimed in claim 5, a capacitance being connected in parallel with the first resistor (R1).

7. The device as claimed in either of claims 5 and 6, it being possible for first resistors (R1) of different magnitudes to be connected in between the inverting input (OP1-) and the output of the first operational amplifier (OP1) for the purpose of setting the current measurement range.

8. The device as claimed in one of the preceding claims, the biochemical molecule to be detected being a nucleic acid and the complementary biochemical molecules being nucleic acids that are complementary to the nucleic acid to be detected.

9. The device as claimed in one of the preceding claims, the potentiostat (P) having a second

operational amplifier (OP2), which is connected as a voltage follower and to whose noninverting input (OP2+) the reference electrode (RE) is connected.

10. The device as claimed in one of the preceding claims, the potentiostat (P) having a third operational amplifier (OP3), to whose output the counterelectrode (GE) is connected and whose inverting input (OP3-) is connected via a second resistor (R2) to the output of the second operational amplifier (OP2) and is connected via a third resistor (R3) to a device for generating a selectable desired voltage, and the noninverting input (OP3+) of the third operational amplifier (OP3) being grounded.

11. The device as claimed in claim 10, a capacitance being connected in between the output of the third operational amplifier (OP3) and the inverting input (OP3-) thereof.

12. A method for the electrochemical detection of at least one type of a biochemical molecule - contained in a liquid - from a group of predetermined biochemical molecules of different types, having the following steps of:

a) providing a means (1) for taking up the liquid, the means (1) having at least one counterelectrode (GE) and a reference electrode (RE) and also more than two working electrodes (AE1, AE2, AE3), at least in each case one working electrode (AE1, AE2, AE3) being provided for the detection of each biochemical molecule, said working electrode being coated with a molecule that is complementary to the biochemical molecule to be detected, so that biochemical molecules of different types can be detected simultaneously,

b) bringing the liquid into contact with the working (AE1, AE2, AE3), counter- (GE) and reference electrodes (RE),
c) simultaneously applying a predetermined voltage profile - which is variable during the measurement - between the working electrodes (AE1, AE2, AE3) and the reference electrode (RE), and
d) measuring the currents flowing through the working electrodes (AE1, AE2, AE3), all of the working electrodes (AE1, AE2, AE3) being held at the same potential during the measurement.

13. The method as claimed in claim 13, the measurement being carried out in parallel or by means of multiplexing.

14. The method as claimed in either of claims 12 and 13, the voltage present between the working electrodes (AE1, AE2, AE3) and the reference electrode (RE) being regulated with a potentiostat (P).